

=> s pseudomonas (w) fluorescens and alginate and mutant
L10 38 PSEUDOMONAS (W) FLUORESCENS AND ALGINATE AND MUTANT

=> s pseudomonas (w) fluorescens and alginate and mutant (w) strain
L11 2 PSEUDOMONAS (W) FLUORESCENS AND ALGINATE AND MUTANT (W) STRAIN

=> d ibib abs 110 1-38

L10 ANSWER 1 OF 38 MEDLINE on STN
ACCESSION NUMBER: 2003447486 MEDLINE
DOCUMENT NUMBER: PubMed ID: 14507360
TITLE: Biofilm formation at the air-liquid interface by the
Pseudomonas fluorescens SBW25 wrinkly
spreader requires an acetylated form of cellulose.
AUTHOR: Spiers Andrew J; Bohannon John; Gehrig Stefanie M; Rainey
Paul B
CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South
Parks Road, Oxford OX1 3RB, UK..
andrew.spiers@plants.ox.ac.uk
SOURCE: Molecular microbiology, (2003 Oct) Vol. 50, No. 1, pp.
15-27.
Journal code: 8712028. ISSN: 0950-382X.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200312
ENTRY DATE: Entered STN: 26 Sep 2003
Last Updated on STN: 18 Dec 2003
Entered Medline: 10 Dec 2003

AB The wrinkly spreader (WS) genotype of Pseudomonas fluorescens SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 2 OF 38 MEDLINE on STN
ACCESSION NUMBER: 2003273771 MEDLINE

DOCUMENT NUMBER: PubMed ID: 12775688
TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.
AUTHOR: Gimmestad Martin; Sletta Havard; Ertesvag Helga; Bakkevig Karianne; Jain Sumita; Suh Sang-jin; Skjak-Braek Gudmund; Ellingsen Trond E; Ohman Dennis E; Valla Svein
CORPORATE SOURCE: Department of Biotechnology, Norwegian University of Science and Technology, Trondheim, Norway.
CONTRACT NUMBER: AI-19146 (NIAID)
SOURCE: Journal of bacteriology, (2003 Jun) Vol. 185, No. 12, pp. 3515-23.
Journal code: 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AF527790
ENTRY MONTH: 200307
ENTRY DATE: Entered STN: 13 Jun 2003
Last Updated on STN: 8 Jul 2003
Entered Medline: 7 Jul 2003

AB Bacterial alginates are produced as 1-4-linked beta-D-mannuronan, followed by epimerization of some of the mannuronic acid residues to alpha-L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose *in vivo* activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 3 OF 38 MEDLINE on STN
ACCESSION NUMBER: 2001654609 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11707327
TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*.
AUTHOR: Morea A; Mathee K; Franklin M J; Giacomini A; O'Regan M; Ohman D E
CORPORATE SOURCE: CRIBI, Biotechnology Centre, University of Padova, Viale G. Colombo 3, 35121 Padova, Italy.
CONTRACT NUMBER: AI-19146 (NIAID)
SOURCE: Gene, (2001 Oct 31) Vol. 278, No. 1-2, pp. 107-14.
Journal code: 7706761. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200201
ENTRY DATE: Entered STN: 15 Nov 2001
Last Updated on STN: 25 Jan 2002
Entered Medline: 10 Jan 2002

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

L10 ANSWER 4 OF 38 MEDLINE on STN
ACCESSION NUMBER: 82144389 MEDLINE
DOCUMENT NUMBER: PubMed ID: 6801192
TITLE: Isolation of alginate-producing mutants of *Pseudomonas fluorescens*, *Pseudomonas putida* and *Pseudomonas mendocina*.
AUTHOR: Govan J R; Fyfe J A; Jarman T R
SOURCE: Journal of general microbiology, (1981 Jul) Vol. 125, No. 1, pp. 217-20.
Journal code: 0375371. ISSN: 0022-1287.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198205
ENTRY DATE: Entered STN: 17 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 21 May 1982

AB Spontaneous alginate-producing (muc) variants were isolated from strains of *Pseudomonas fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 10⁸ by selecting for carbenicillin resistance. The infrared spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii* and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteroni*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 5 OF 38 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved. (2006) on STN

ACCESSION NUMBER: 93:76265 AGRICOLA
DOCUMENT NUMBER: IND93050364
TITLE: Influence of *Pseudomonas fluorescens* on hyphal growth and biocontrol activity of *Trichoderma harizanum* in the rhizosphere and rhizosphere of pea.

AUTHOR(S): Dandurand, L.M.; Knudsen, G.R.
AVAILABILITY: DNAL (464.8 P56)
SOURCE: *Phytopathology*, Mar 1993. Vol. 83, No. 3. p. 265-270
Publisher: St. Paul, Minn. : American
Phytopathological Society.
CODEN: PHYTAJ; ISSN: 0031-949X
NOTE: Includes references.
DOCUMENT TYPE: Article
FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension
LANGUAGE: English

AB *Trichoderma harzianum* isolate ThzID1 was grown in liquid culture, was formulated with alginate and polyethylene glycol 8000; and was milled into fine granules (average diameter 500 micrometers). Granules contained chlamydospores, conidia, and hyphal fragments. Viability of the encapsulated fungus remained high for at least 6 mo when stored at 5 C (i.e., > 90% of the granules produced hyphal growth when incubated on agar); viability was reduced significantly when granules were stored at 22 C. Application of the granular formulation of *T. harzianum* to pea seeds reduced root rot by *Aphanomyces euteiches* f. sp. *pisi* in growth-chamber experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of *Pseudomonas fluorescens* strain 2-79RN(10), which produces a phenazine antibiotic, also reduced *Aphanomyces* root rot but to a lesser extent than did *T. harzianum* ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of *T. harzianum* and 2-79RN(10) compared to treatment with *T. harzianum* alone. Root rot was not reduced by the mutant *P. fluorescens* strain 2-79-B46, which lacks phenazine. Treatment with *T. harzianum* plus 2-79-B46 resulted in the same level of disease control achieved by *T. harzianum* alone. These results suggest that the biocontrol mechanism of *P. fluorescens* 2-79RN(10) neither inhibited nor enhanced the biocontrol activity of *T. harzianum* ThzID1. In other experiments, density of *T. harzianum* hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN(10), but when 2-79-B46 was added, density was greater after 5 days. The colony radius of *T. harzianum* was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of *T. harzianum* in the presence of bacteria was not the direct result of stimulation of seed exudation by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 6 OF 38 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved.
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ACCESSION NUMBER: 91:81096 AGRICOLA
DOCUMENT NUMBER: IND91045062
TITLE: Influence of an antagonistic strain of *Pseudomonas fluorescens* on growth and ability of *Trichoderma harzianum* to colonize sclerotia of *Sclerotinia sclerotiorum* in soil.

AUTHOR(S): Bin, L.; Knudsen, G.R.; Eschen, D.J.
CORPORATE SOURCE: University of Idaho, Moscow
AVAILABILITY: DNAL (464.8 P56)
SOURCE: *Phytopathology*, Sept 1991. Vol. 81, No. 9. p. 994-1000
Publisher: St. Paul, Minn. : American
Phytopathological Society.
CODEN: PHYTAJ; ISSN: 0031-949X
NOTE: Includes references.
DOCUMENT TYPE: Article

FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension
LANGUAGE: English
AB *Pseudomonas fluorescens* strain 2-79RN10 (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus *Trichoderma harzianum* isolate THzID1, which was formulated into alginic acid pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79RN10 maintained its initial high populations (approximately 3×10^{14} or 3×10^{15} cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79RN10 decreased by approximately four log₁₀ units over a 3-wk period, and did not affect the ability of *Trichoderma* spp. to colonize sclerotia of *Sclerotinia sclerotiorum*. In two years of field experiments using raw or steamed soil in microplots, populations of 2-79RN10 decreased gradually after 1-2 wk and did not reduce the ability of *Trichoderma* spp. to colonize sclerotia of *S. sclerotiorum*. Colonization of sclerotia by *Trichoderma* spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%) when THzID1 was added, suggesting possible inhibition of THzID1 by indigenous soil microbes, or utilization by ThzID1 of nutrients released by steaming of soil. In treatments where ThzID1 was not added, low levels of colonization of sclerotia were observed, apparently due to indigenous *Trichoderma* spp., and these levels were higher in raw soil (mean = 18%) than in steamed soil (mean = 5%). These results suggest that under certain restrictive conditions, high population levels of antagonistic bacteria in bulk soil suppressed a fungal biocontrol agent, but that this suppressive effect was reduced or eliminated when a high bacterial population was present.

L10 ANSWER 7 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 2003:510388 BIOSIS

DOCUMENT NUMBER: PREV200300497223

TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose.

AUTHOR(S): Spiers, Andrew J. [Reprint Author]; Bohannon, John; Gehrig, Stefanie M.; Rainey, Paul B.

CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South Parks Road, Oxford, OX1 3RB, UK
andrew.spiers@plants.ox.ac.uk

SOURCE: Molecular Microbiology, (October 2003) Vol. 50, No. 1, pp. 15-27. print.
ISSN: 0950-382X (ISSN print).

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 29 Oct 2003

Last Updated on STN: 29 Oct 2003

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFHI from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginic polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its

similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 8 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
ACCESSION NUMBER: 2003:303128 BIOSIS

DOCUMENT NUMBER: PREV200300303128

TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.

AUTHOR(S): Gimmestad, Martin; Sletta, Havard; Ertesvag, Helga; Bakkevig, Karianne; Jain, Sumita; Suh, Sang-Jin; Skjak-Braek, Gudmund; Ellingsen, Trond E.; Ohman, Dennis E.; Valla, Svein [Reprint Author]

CORPORATE SOURCE: Department of Biotechnology, NTNU Norwegian University of Science and Technology, N-7491, Trondheim, Norway
svein.valla@biotech.ntnu.no

SOURCE: Journal of Bacteriology, (June 2003) Vol. 185, No. 12, pp. 3515-3523. print.

CODEN: JOBAAY. ISSN: 0021-9193.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 2 Jul 2003

Last Updated on STN: 2 Jul 2003

AB Bacterial alginates are produced as 1-4-linked beta-D-mannuronan, followed by epimerization of some of the mannuronic acid residues to alpha-L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose *in vivo* activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 9 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
ACCESSION NUMBER: 2002:41402 BIOSIS

DOCUMENT NUMBER: PREV200200041402

TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*.

AUTHOR(S): Morea, Antonella; Mathee, Kalai; Franklin, Michael J.;

Giacomini, Alessio; O'Regan, Michael; Ohman, Dennis E.
[Reprint author]

CORPORATE SOURCE: Department of Microbiology and Immunology, Medical College, Virginia Commonwealth University, 1101 E. Marshall Street, Virginia Campus, 5-047 Sanger Hall, Richmond, VA, 23298-0678, USA
deohman@hsc.vcu.edu

SOURCE: Gene (Amsterdam), (31 October, 2001) Vol. 278, No. 1-2, pp. 107-114. print.

CODEN: GENED6. ISSN: 0378-1119.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 2 Jan 2002
Last Updated on STN: 25 Feb 2002

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

L10 ANSWER 10 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1993:233118 BIOSIS

DOCUMENT NUMBER: PREV199395124293

TITLE: Influence of *Pseudomonas fluorescens* on hyphal growth and biocontrol activity of *Trichoderma harzianum* in the spermosphere and rhizosphere of pea.

AUTHOR(S): Dandurand, L. M.; Knudsen, G. R.

CORPORATE SOURCE: Plant Pathol. Div., Univ. Idaho, Moscow, ID 83843, USA

SOURCE: Phytopathology, (1993) Vol. 83, No. 3, pp. 265-270.

CODEN: PHYTAJ. ISSN: 0031-949X.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 7 May 1993
Last Updated on STN: 7 May 1993

AB *Trichoderma harzianum* isolate ThzID1 was grown in liquid culture, was formulated with alginate and polyethylene glycol 18000, and was milled into fine granules (average diameter 500 μ m). Granules contained chlamydospores, conidia, and hyphal fragments. Viability of the encapsulated fungus remained high for at least 6 mo when stored at 5 C (i.e., > 90% of the granules produced hyphal growth when incubated on agar); viability was reduced significantly when granules were stored at 22 C. Application of the granular formulation of *T. harzianum* to pea seeds reduced root rot by *Aphanomyces euteiches* f. sp. *pisi* in growth-chamber

experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of *Pseudomonas fluorescens* strain 2-79RN-10, which produces a phenazine antibiotic, also reduced *Aphanomyces* root rot but to a lesser extent than did *T. harzianum* ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of *T. harzianum* and 2-79RN-10 compared to treatment with *T. harzianum* alone. Root rot was not reduced by the mutant *P. fluorescens* strain 2-79-B46, which lacks phenazine. Treatment with *T. harzianum* plus 2-79-B46 resulted in the same level of disease control achieved by *T. harzianum* alone. These results suggest that the biocontrol mechanism of *P. fluorescens* 2-79RN-10 neither inhibited nor enhanced the biocontrol activity of *T. harzianum* ThzID1. In other experiments, density of *T. harzianum* hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN-10, but when 2-79-B46 was added, density was greater after 5 days. The colony radius of *T. harzianum* was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of *T. harzianum* in the presence of bacteria was not the direct result of stimulation of seed germination by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 11 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1992:361205 BIOSIS
DOCUMENT NUMBER: PREV199243039355; BR43:39355
TITLE: BIOCONVERSION OF VANILLIN INTO VANILLIC ACID BY PSEUDOMONAS-FLUORESCENS STRAIN BTP9 CELL REACTORS AND MUTANTS STUDY.
AUTHOR(S): BARE G [Reprint author]; GERARD J; JACQUES P; DELAUNOIS V; THONART P
CORPORATE SOURCE: CENTRE WALLON BIOL IND, UNIV LIEGE, FSA GX, SART-TILMAN B40, 4000 LIEGE, BELG
SOURCE: Applied Biochemistry and Biotechnology, (1992) Vol. 34-35, pp. 499-514.
Meeting Info.: THIRTEENTH SYMPOSIUM ON BIOTECHNOLOGY FOR FUELS AND CHEMICALS, COLORADO SPRINGS, COLORADO, USA, MAY 6-10, 1991. APPL BIOCHEM BIOTECHNOL.
CODEN: ABIBDL. ISSN: 0273-2289.
DOCUMENT TYPE: Conference; (Meeting)
FILE SEGMENT: BR
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 30 Jul 1992
Last Updated on STN: 30 Jul 1992

L10 ANSWER 12 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1991:482821 BIOSIS
DOCUMENT NUMBER: PREV199192116581; BA92:116581
TITLE: INFLUENCE OF AN ANTAGONISTIC STRAIN OF PSEUDOMONAS-FLUORESCENS ON GROWTH AND ABILITY OF TRICHODERMA-HARZIANUM TO COLONIZE SCLEROTIA OF SCLEROTINIA-SCLEROTIORUM IN SOIL.
AUTHOR(S): BIN L [Reprint author]; KNUDSEN G R; ESCHEN D J
CORPORATE SOURCE: PLANT PATHOL DIV, DEP PLANT SOIL AND ENTOMOLOGICAL SCI, UNIV IDAHO, MOSCOW, IDAHO 83843, USA
SOURCE: Phytopathology, (1991) Vol. 81, No. 9, pp. 994-1000.
CODEN: PHYTAJ. ISSN: 0031-949X.
DOCUMENT TYPE: Article
FILE SEGMENT: BA

LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 26 Oct 1991
Last Updated on STN: 26 Oct 1991
AB Pseudomonas fluorescens strain 2-79N10 (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus Trichoderma harzianum isolate ThzID1, which was formulated into alginate pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79RN10 maintained its initial high populations (approximately 3 + 104 or 3 + 107 cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79RN10 decreased by approximately four log₁₀ units over a 3-wk period, and did not affect the ability of Trichoderma spp. to colonize sclerotinia of Sclerotinia sclerotiorum. In two years of field experiments using a raw or steamed soil in microplots, populations of 2-79RN10 decreased gradually after 1-2 wk and did not reduce the ability of Trichoderma spp. to colonize sclerotinia S. sclerotiorum. Colonization of sclerotinia by Trichoderma spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%) when ThzID1 was added, suggesting possible inhibition of ThzID1 by indigenous soil microbes, or utilization by ThzID1 of nutrients released by steaming of soil. In treatments where ThzID1 was not added, low levels of colonization of sclerotinia were observed, apparently due to indigenous Trichoderma spp., and these levels were higher in raw soil (mean = 18%) than in steamed soil (mean = 5%). These results suggest that under certain restrictive conditions, high population levels of antagonistic bacteria in bulk soil suppressed a fungal biocontrol agent, but that this suppressive effect was reduced or eliminated when a high bacterial population was not present.

L10 ANSWER 13 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1984:195488 BIOSIS
DOCUMENT NUMBER: PREV198477028472; BA77:28472
TITLE: SELECTION OF ATTACHMENT MUTANTS DURING THE
CONTINUOUS CULTURE OF PSEUDOMONAS-
FLUORESCENS AND RELATIONSHIP BETWEEN ATTACHMENT
ABILITY AND SURFACE COMPOSITION.
AUTHOR(S): PRINGLE J H [Reprint author]; FLETCHER M; ELLWOOD D C
CORPORATE SOURCE: DEP OF MOLECULAR BIOL, UNIV OF EDINBURGH, EDINBURGH EH9
3JR, UK
SOURCE: Journal of General Microbiology, (1983) Vol. 129, No. 8,
pp. 2557-2570.
CODEN: JGMIAN. ISSN: 0022-1287.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH

AB A P. fluorescens strain, isolated from a freshwater source on a plastic substratum, was grown in continuous culture in minimal medium. The adsbubble (adsorptive bubble separation) process foam-fractionated, wild-type cells from the fermenter during flow conditions. This selection pressure favored the enrichment of 2 major classes of mutant, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide. A mucoid mutant, found predominantly in the aqueous phase, produced an alginate expolymer. The 2nd class of mutant was isolated from the walls of the fermenter and, like the wild-type, produced little expolymer. This mutant, with crenated colony morphology, showed increased attachment to solid surfaces compared to the wild-type and mucoid cells when assayed for attachment to polystyrene surfaces for 2 h. Outer-membrane protein, lipopolysaccharides, and expolysaccharides of the wild-type and both

mutants were analyzed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to micro-environments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 14 OF 38 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

ACCESSION NUMBER: 1982:178834 BIOSIS

DOCUMENT NUMBER: PREV198273038818; BA73:38818

TITLE: ISOLATION OF ALGINATE PRODUCING MUTANTS OF PSEUDOMONAS-FLUORESCENS PSEUDOMONAS-PUTIDA AND PSEUDOMONAS-MENDOCINA.

AUTHOR(S): GOVAN J R W [Reprint author]; FYFE J A M; JARMAN T R

CORPORATE SOURCE: DEP OF BACTERIOLOGY, UNIV OF EDINBURGH, MED SCHOOL, TEVIOT PLACE, EDINBURGH EH8 9AG, UK

SOURCE: Journal of General Microbiology, (1981) Vol. 125, No. 1, pp. 217-220.

CODEN: JGMIAN. ISSN: 0022-1287.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

AB Spontaneous alginate-producing (muc) variants were isolated from strains of *P. fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 10⁸ by selecting for carbenicillin resistance. The IR spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii* and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteroni*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 15 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:101282 CAPLUS

DOCUMENT NUMBER: 140:162433

TITLE: Engineering *Pseudomonas fluorescens* variants for improved alginate production for use in cosmetics, pharmaceuticals and nutrients and animal feed

INVENTOR(S): Gimmestad, Martin; Sletta, Havard; Karunakaran, Karuna Ponniah; Bakkevig, Karianne; Ertesvag, Helga; Ellingsen, Trond; Skjak-Braek, Gudmund; Valla, Svein

PATENT ASSIGNEE(S): FMC Biopolymer AS, Norway

SOURCE: PCT Int. Appl., 76 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004011628	A1	20040205	WO 2003-N0257	20030724
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
CA 2493638	AA	20040205	CA 2003-2493638	20030724
AU 2003248516	A1	20040216	AU 2003-248516	20030724
EP 1543105	A1	20050622	EP 2003-771512	20030724

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
CN 1685037	A	20051019	CN 2003-822409	20030724
JP 2005533515	T2	20051110	JP 2004-524404	20030724
NO 2005000480	A	20050408	NO 2005-480	20050127
US 2006063237	A1	20060323	US 2005-522510	20050917
PRIORITY APPLN. INFO.:			NO 2002-3581	A 20020726
			WO 2003-NO257	W 20030724

AB It is described biol. pure bacterial cultures of mutant strains of *Pseudomonas fluorescens*, which produces large amts. of alginic acid. The alginic acid may contain a certain determined content of manuronate and guluronate residues, possible presence and determined level of acetyl groups in the alginic acid, and a desired mol. weight of the alginic acid. Also high yielding mutants with regulation of alginic acid production, is described. The invention further provides methods for producing new mutant strains of *Pseudomonas fluorescens* and variants thereof, and use of the resulting strains in improved alginic acid prodn for use in drugs, cosmetics, animal feed and nutrients.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 16 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN
 ACCESSION NUMBER: 2003:817271 CAPLUS
 DOCUMENT NUMBER: 140:25307
 TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose
 AUTHOR(S): Spiers, Andrew J.; Bohannon, John; Gehrig, Stefanie M.; Rainey, Paul B.
 CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, Oxford, OX1 3RB, UK
 SOURCE: Molecular Microbiology (2003), 50(1), 15-27
 CODEN: MOMIEE; ISSN: 0950-382X
 PUBLISHER: Blackwell Publishing Ltd.
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overprodn. of a cellulosic polymer, the product of the wss operon. Chemical anal. of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* anal. of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologs of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginic acid polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to Mind-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibers, sheets and clumped material. Quant. analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered phys. properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated

cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

REFERENCE COUNT: 52 THERE ARE 52 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 17 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 2003:466304 CAPLUS
DOCUMENT NUMBER: 139:335185
TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation
AUTHOR(S): Gimmestad, Martin; Sletta, Havard; Ertesvag, Helga; Bakkevig, Karianne; Jain, Sumita; Suh, Sang-jin; Skjok-Braek, Gudmund; Ellingsen, Trond E.; Ohman, Dennis E.; Valla, Svein
CORPORATE SOURCE: Department of Biotechnology, Norwegian University of Science and Technology, Trondheim, Norway
SOURCE: Journal of Bacteriology (2003), 185(12), 3515-3523
CODEN: JOBAAY; ISSN: 0021-9193
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose *in vivo* activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate mols.: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.
REFERENCE COUNT: 51 THERE ARE 51 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 18 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 2001:826434 CAPLUS
DOCUMENT NUMBER: 136:364494
TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*
AUTHOR(S): Morea, Antonella; Mathee, Kalai; Franklin, Michael J.; Giacomini, Alessio; O'Regan, Michael; Ohman, Dennis E.
CORPORATE SOURCE: Biotechnology Centre, CRIBI, University of Padova, Padua, 35121, Italy
SOURCE: Gene (2001), 278(1-2), 107-114
CODEN: GENED6; ISSN: 0378-1119
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of

genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approx. in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained 1-guluronate as determined by proton NMR spectroscopy. A sequence anal. of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 19 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1995:833235 CAPLUS

DOCUMENT NUMBER: 123:220822

TITLE: *Pseudomonas fluorescens* mutant strains for the biocontrol of plant pathogenic fungi

INVENTOR(S): Lam, Stephen; Torkewitz, Nancy

PATENT ASSIGNEE(S): Ciba-Geigy A.-G., Switz.

SOURCE: PCT Int. Appl., 38 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9520040	A1	19950727	WO 1995-IB23	19950111
W: AM, AU, BB, BG, BR, BY, CA, CN, CZ, EE, FI, GE, HU, JP, KG, KP, KR, KZ, LK, LR, LT, LV, MD, MG, MN, MX, NO, NZ, PL, RO, RU, SI, SK, TJ, TT, UA, US, UZ, VN				
RW: KE, MW, SD, SZ, AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
US 5496547	A	19960305	US 1994-185623	19940124
AU 9512789	A1	19950808	AU 1995-12789	19950111
AU 694923	B2	19980806		
EP 743980	A1	19961127	EP 1995-903897	19950111
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
CN 1139453	A	19970101	CN 1995-191311	19950111
JP 09508269	T2	19970826	JP 1995-519443	19950111
RU 2154943	C2	20000827	RU 1996-117351	19950111
PRIORITY APPLN. INFO.:			US 1994-185623	A2 19940124
			WO 1995-IB23	W 19950111

AB Mutant strains of *Pseudomonas* were isolated by transposon insertion mutation and screening which have enhanced biocontrol properties, particularly against *Rhizoctonia solani*. A plasmid pCIB116 (containing a Tn5-promoterless lac transposable element) suitable for transposon mutagenesis was constructed and transferred to *Pseudomonas*

strain CGA 267356 to generate a collection of insertion mutants. About 10,000 mutants were tested for their ability to inhibit growth of *Neurospora* in vitro, and selected strains were further tested for their ability to control infestation by *R. solani* of cotton in greenhouse tests. Two insertion mutants provided better disease control against *R. solani* than did wild-type strains. Mutant strains CGA 319115 and CGA 32170 were applied as granules consisting of a finely divided carrier (e.g., vermiculite) and a polymer layer layer, wherein the polymer is (a) a film-forming, water-soluble, and essentially non-crosslinked polymer or (b) a film-forming, structurally crosslinked, water-swellable polysaccharide (e.g., κ -carrageenan or sodium alginate).

L10 ANSWER 20 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1992:406044 CAPLUS
DOCUMENT NUMBER: 117:6044
TITLE: Bioconversion of vanillin into vanillic acid by *Pseudomonas fluorescens* strain BTP9: cell reactors and mutants study
AUTHOR(S): Bare, G.; Gerard, J.; Jacques, P.; Delaunois, V.; Thonart, P.
CORPORATE SOURCE: Cent. Wallon Biol. Ind., Univ. Liege, Liege, 4000, Belg.
SOURCE: Applied Biochemistry and Biotechnology (1992), 34-35, 499-510
CODEN: ABIBDL; ISSN: 0273-2289
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The ability of a fluorescent *Pseudomonas* to bioconvert vanillin, a phenolic compound, to vanillic acid was investigated. Free and immobilized cell reactors were tested. With free cells, the optimal yield reaches 98% after 6.5 h of bioconversion. With cells immobilized in alginate beads, transformation rate is only 47% after 13 h of conversion. Nevertheless, a continuous immobilized cell reactor was used for 76 h. With this, the optimal yield is >80%. The effects of residence time and cell concentration of the alginate beads in the reactor over the reactor's productivity also were studied. Catabolically blocked mutants for vanillic acid degradation were searched. To screen these mutants, a new and very sensitive method was developed. The results of mutant screenings are discussed.

L10 ANSWER 21 OF 38 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1983:536524 CAPLUS
DOCUMENT NUMBER: 99:136524
TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas fluorescens* and relationship between attachment ability and surface composition
AUTHOR(S): Pringle, J. Howard; Fletcher, Madilyn; Ellwood, D. C.
CORPORATE SOURCE: Dep. Environ. Sci., University of Warwick, Coventry, CV4 7AL, UK
SOURCE: Journal of General Microbiology (1983), 129(8), 2557-69
CODEN: JGMIAN; ISSN: 0022-1287
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A strain of *P. fluorescens* that had been isolated from a freshwater source on a plastic substratum was grown in continuous culture in minimal medium. The adsorptive bubble separation process foam-fractionated wild-type cells from the fermentor during flow conditions. This selection pressure favored the enrichment of 2 major classes of mutants, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide, whereas a mucoid mutant, found predominantly in the aqueous phase, produced an

alginate exopolymer. The 2nd class of mutant was isolated from the walls of the fermentor and, like the wild-type, produced little exopolymer. This mutant, with crenated colony morphol., showed increased attachment to solid surfaces compared to the wild-type and mucoid cells when assayed for attachment to polystyrene surfaces for 2 h. Outer-membrane protein, lipopolysaccharides, and exopolysaccharides of the wild-type and both mutants were analyzed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to microenvironments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 22 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2003:107538 LIFESCI

TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose

AUTHOR: Spiers, A.J.; Bohannon, J.; Gehrig, S.M.; Rainey, P.B.

CORPORATE SOURCE: Department of Plant Sciences, University of Oxford, South Parks Road, Oxford OX1 3RB, UK.; E-mail: andrew.spiers@plants.ox.ac.uk

SOURCE: Molecular Microbiology [Mol. Microbiol.], (2003)1000) vol. 50, no. 1, pp. 15-27.

ISSN: 0950-382X.

DOCUMENT TYPE: Journal

FILE SEGMENT: G; J

LANGUAGE: English

SUMMARY LANGUAGE: English

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHJI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHJI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 23 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 2003:73780 LIFESCI

TITLE: The *Pseudomonas fluorescens* AlgG Protein, but Not Its Mannuronan C-5- Epimerase Activity, Is Needed for Alginate Polymer Formation

AUTHOR: Gimmestad, M.; Sletta, H.; Ertesvaag, H.; Bakkevig, K.;

CORPORATE SOURCE: Jain, S.; Suh, S.; Skjaak-Braek, G.; Ellingsen, T.E.; Ohman, D.E.; Valla, S.*
Department of Biotechnology, NTNU Norwegian University of Science and Technology, N-7491 Trondheim, Norway; E-mail: svein.valla@biotech.ntnu.no

SOURCE: Journal of Bacteriology [J. Bacteriol.], (20030600) vol. 185, no. 12, pp. 3515-3523.
ISSN: .0021-9193.

DOCUMENT TYPE: Journal
FILE SEGMENT: G; J
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Bacterial alginates are produced as 1-4-linked beta-D-mannuronan, followed by epimerization of some of the mannuronic acid residues to [alpha] -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4- enepyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose in vivo activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginate molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 24 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN
ACCESSION NUMBER: 2002:23850 LIFESCI
TITLE: Characterization of algG encoding C5-epimerase in the alginate biosynthetic gene cluster of *Pseudomonas fluorescens*
AUTHOR: Morea, A.; Mathee, K.; Franklin, M.J.; Giacomini, A.; O'Regan, M.; Ohman, D.E.
CORPORATE SOURCE: CRIBI, Biotechnology Centre, University of Padova, Viale G. Colombo 3, 35121 Padova Italy
SOURCE: Gene, (20011031) vol. 278, no. 1-2, pp. 107-114.
ISSN: 0378-1119.
DOCUMENT TYPE: Journal
FILE SEGMENT: G; J
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The organization of the alginate gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginate biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginate biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginate as polymannuronate due to its C5-epimerase defect, complementation was observed and the alginate from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that

were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginate production, suggesting a potential role for this protein in polymer formation.

L10 ANSWER 25 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:90362 LIFESCI

TITLE: Influence of *Pseudomonas fluorescens*

on hyphal growth and biocontrol activity of *Trichoderma harzianum* in the spermosphere and rhizosphere of pea.

AUTHOR: Dandurand, L.M.; Knudsen, G.R.

CORPORATE SOURCE: Plant Pathol. Div., Univ. Idaho, Moscow, ID 83843, USA

SOURCE: PHYTOPATHOLOGY., (1993) vol. 83, no. 3, pp. 265-270.

ISSN: 0031-949X.

DOCUMENT TYPE: Journal

FILE SEGMENT: A; K; J

LANGUAGE: English

SUMMARY LANGUAGE: English

AB *Trichoderma harzianum* isolate ThzID1 was grown in liquid culture, was formulated with alginate and polyethylene glycol 8000, and was milled into fine granules (average diameter 500 μ m). Granules contained chlamydospores, conidia, and hyphal fragments. Viability of the encapsulated fungus remained high for at least 6 mo when stored at 5 C (i.e., >90% of the granules produced hyphal growth when incubated on agar); viability was reduced significantly when granules were stored at 22 C. Application of the granular formulation of *T. harzianum* to pea seeds reduced root rot by *Aphanomyces euteiches* f. sp. *pisi* in growth-chamber experiments and also increased plant top weights compared to noncoated seeds. Seed treatment with slurries of *Pseudomonas fluorescens* strain 2-79RN sub(10), which produces a phenazine antibiotic, also reduced *Aphanomyces* root rot but to a lesser extent than did *T. harzianum* ThzID1. Disease suppression was not significantly different when seeds were treated with a combination of *T. harzianum* and 2-79RN sub(10) compared to treatment with *T. harzianum* alone. Root rot was not reduced by the mutant *P. fluorescens* strain 2-79-B46, which lacks phenazine. Treatment with *T. harzianum* plus 2-79-B46 resulted in the same level of disease control achieved by *T. harzianum* alone. These results suggest that the biocontrol mechanism of *P. fluorescens* 2-79RN sub(10) neither inhibited nor enhanced the biocontrol activity of *T. harzianum* ThzID1. In other experiments, density of *T. harzianum* hyphae originating from coated pea seeds in soil was not affected by the addition of 2-79RN sub(10), but when 2-79-B46 was added, density was greater after 5 days. The colony radius of *T. harzianum* was initially enhanced (at 3 days) by the addition of either strain, but the effect diminished by day 5. The same treatments were then applied to peas and to glass beads of equivalent size, and similar effects of the added bacterial strains were observed on both substrates, suggesting that the growth enhancement of *T. harzianum* in the presence of bacteria was not the direct result of stimulation of seed exudation by the bacteria. Our results provide a potentially improved formulation methodology for coating seeds with biocontrol organisms and methods for evaluating the compatibility of fungal and bacterial biocontrol agents applied to seeds.

L10 ANSWER 26 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:7774 LIFESCI

TITLE: Bioconversion of vanillin into vanillic acid by

Pseudomonas fluorescens strain BTP9:

Cell reactors and mutants study.

PROCEEDINGS OF THE THIRTEENTH SYMPOSIUM ON BIOTECHNOLOGY

FOR FUELS AND CHEMICALS.
AUTHOR: Bare, G.; Gerard, J.; Jacques, P.; Delaunois, V.; Thonart, P.
CORPORATE SOURCE: Cent. Wallon Biol. Ind., Univ. Liege, F.S.A. Gx, Sart-Tilman, B40, 4000 Liege, Belgium
SOURCE: APPL. BIOCHEM. BIOTECHNOL., (1992) pp. 499-512.
Meeting Info.: 13. Symposium on Biotechnology for Fuels and Chemicals. (np).
DOCUMENT TYPE: Book
TREATMENT CODE: Conference
FILE SEGMENT: A; W
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The ability of a fluorescent *Pseudomonas* to bioconvert vanillin, a phenolic compound, into vanillic acid was investigated. Free and immobilized cell reactors were tested. With free cells, the optimal yield reaches 98% after 6 h and 30 min of bioconversion. With cells immobilized in alginate beads, transformation rate is only 47% after 13 h of conversion. Nevertheless, a continuous immobilized cell reactor was used for 76 h. With this one, the optimal yield is higher than 80%. The influence of the residence time and cell concentration of the alginate beads in the reactor over the reactor's productivity has also been studied. Catabolically blocked mutants for vanillic acid degradation were searched.

L10 ANSWER 27 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 93:5127 LIFESCI
TITLE: Influence of an antagonistic strain of *Pseudomonas fluorescens* on growth and ability of *Trichoderma harzianum* to colonize sclerotinia of *Sclerotinia sclerotiorum* in soil.
AUTHOR: Bin, L.; Knudsen, G.R.; Eschen, D.J.
CORPORATE SOURCE: Plant Pathol. Div., Dep. Plant, Soil, and Entomol. Sci., Univ. Idaho, Moscow, ID 83843, USA
SOURCE: PHYTOPATHOLOGY., (1991) vol. 81, no. 9, pp. 994-1000.
DOCUMENT TYPE: Journal
FILE SEGMENT: J; A; W; K
LANGUAGE: English
SUMMARY LANGUAGE: English

AB *Pseudomonas fluorescens* strain 2-79RN sub(10) (nalidixic acid and rifampicin-resistant mutant of wild type strain 2-79) was used to study potential effects of bacterial antagonism in soil on growth and biocontrol efficacy of the biocontrol fungus *Trichoderma harzianum* isolate ThzID1, which was formulated into alginate pellets. In steamed soil (25 C, -100 or -500 kPa matric potential), strain 2-79RN sub(10) maintained its initial high populations (approximately 3 X 10⁴ or 3 X 10⁷ cfu/g of soil) over a 14-day period, and significantly reduced hyphal radius, hyphal density, and recoverable numbers of propagules of ThzID1. In raw soil under similar environmental conditions (22-25 C, -10 to -1,000 kPa), populations of 2-79RN sub(10) decreased by approximately four log sub(10) units over a 3-wk period, and did not affect the ability of *Trichoderma* spp. to colonize sclerotinia of *Sclerotinia sclerotiorum*. Populations of 2-79RN sub(10) decreased gradually after 1-2 wk and did not reduce the ability of *Trichoderma* spp. to colonize sclerotinia of *S. sclerotiorum*. Colonization of sclerotia by *Trichoderma* spp. after 9 wk was significantly higher in steamed soil (mean = 65%) than in raw soil (mean = 30%).

L10 ANSWER 28 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN

ACCESSION NUMBER: 83:41158 LIFESCI
TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas fluorescens* and relationship between attachment ability and surface composition.

AUTHOR: Pringle, J.H.; Fletcher, M.; Ellwood, D.C.
CORPORATE SOURCE: Dep. Environ. Sci., Univ. Warwick, Coventry CV4 7AL, UK
SOURCE: J. GEN. MICROBIOL., (1983) vol. 129, no. 8, pp. 2557-2569.
DOCUMENT TYPE: Journal
FILE SEGMENT: J; M
LANGUAGE: English
SUMMARY LANGUAGE: English

AB A strain of *P. fluorescens* that had been isolated from a freshwater source on a plastic substratum was grown in continuous culture in minimal medium. The "adsbubble" process (adsorptive bubble separation process) was found to foam-fractionate wild-type cells from the fermenter during flow conditions. This selection pressure favoured the enrichment of two major classes of mutant, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide, whereas a "mucoid" mutant, found predominantly in the aqueous-phase, produced an alginate exopolymer. Outer-membrane protein, lipopolysaccharides and exopolysaccharides of the wild-type and both mutants were analysed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to micro-environments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 29 OF 38 LIFESCI COPYRIGHT 2006 CSA on STN
ACCESSION NUMBER: 81:18399 LIFESCI
TITLE: Isolation of Alginate-Producing Mutants
of *Pseudomonas fluorescens*, *Pseudomonas putida* and *Pseudomonas mendocina*.
AUTHOR: Govan, J.R.W.; Fyfe, J.A.M.; Jarman, T.R.
CORPORATE SOURCE: Dept. Bacteriol., Univ. Edinburgh, Med. Sch., Teviot Place,
Edinburgh EH8 9AG, UK
SOURCE: J. GEN. MICROBIOL., (1981) vol. 125, no. 1, pp. 217-220.
DOCUMENT TYPE: Journal
FILE SEGMENT: J
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Spontaneous alginate-producing (muc) variants were isolated from strains of *P. fluorescens*, *P. putida* and *P. mendocina* at a frequency of 1 in 10 super(8) by selecting for carbenicillin resistance. The infrared spectrum of the bacterial exopolysaccharide was typical of an acetylated alginate similar to that previously described in *Azotobacter vinelandii* and in mucoid variants of *P. aeruginosa*. Mucoid variants were not isolated from *P. stutzeri*, *P. pseudoalcaligenes*, *P. testosteronei*, *P. diminuta*, *P. acidovorans*, *P. cepacia* or *P. maltophilia*.

L10 ANSWER 30 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2003:37222783 BIOTECHNO
TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose
AUTHOR: Spiers A.J.; Bohannon J.; Gehrig S.M.; Rainey P.B.
CORPORATE SOURCE: A.J. Spiers, Department of Plant Sciences, University of Oxford, South Parks Road, Oxford OX1 3RB, United Kingdom.
E-mail: andrew.spiers@plants.ox.ac.uk
SOURCE: Molecular Microbiology, (2003), 50/1 (15-27), 52 reference(s)
DOCUMENT TYPE: CODEN: MOMIEE ISSN: 0950-382X
FILE SEGMENT: Journal; Article
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 2003:37222783 BIOTECHNO
AB The wrinkly spreader (WS) genotype of *Pseudomonas*

fluorescens SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 31 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2003:36676417 BIOTECHNO
TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation
AUTHOR: Gimmestad M.; Sletta H.; Ertesvag H.; Bakkevig K.; Jain S.; Suh S.-J.; Skjak-Braek G.; Ellingsen T.E.; Ohman D.E.; Valla S.
CORPORATE SOURCE: S. Valla, Department of Biotechnology, NTNU Norwegian Univ. Sci./Technol., N-7491 Trondheim, Norway.
E-mail: svein.valla@biotech.ntnu.no
SOURCE: Journal of Bacteriology, (2003), 185/12 (3515-3523), 51 reference(s)
CODEN: JOBAAY ISSN: 0021-9193
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 2003:36676417 BIOTECHNO
AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL, whose *in vivo* activity is much more limited in the presence of

AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginic molecules: one class being pure mannuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

L10 ANSWER 32 OF 38 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2001:33055784 BIOTECHNO
TITLE: Characterization of algG encoding C5-epimerase in the alginic biosynthetic gene cluster of *Pseudomonas fluorescens*
AUTHOR: Morea A.; Mathee K.; Franklin M.J.; Giacomini A.; O'Regan M.; Ohman D.E.
CORPORATE SOURCE: D.E. Ohman, Department of Microbiology, M.C.V.C. Virginia Commonwealth Univ., 5-047 Sanger Hall, 1101 E. Marshall Street, Richmond, VA 23298-0678, United States.
E-mail: deohman@hsc.vcu.edu
SOURCE: Gene, (31 OCT 2001), 278/1-2 (107-114), 33 reference(s)
CODEN: GENED6 ISSN: 0378-1119
PUBLISHER ITEM IDENT.: S0378111901006850
DOCUMENT TYPE: Journal; Article
COUNTRY: Netherlands
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 2001:33055784 BIOTECHNO
AB The organization of the alginic gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginic biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginic biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginic acid polymannuronate due to its C5-epimerase defect, complementation was observed and the alginic acid from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginic acid production, suggesting a potential role for this protein in polymer formation. .COPYRGT. 2001 Elsevier Science B.V. All rights reserved.

L10 ANSWER 33 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN
ACCESSION NUMBER: 2005568101 EMBASE
TITLE: Role of the *Pseudomonas fluorescens* alginate lyase (AlgL) in clearing the periplasm of alginates not exported to the extracellular environment.
AUTHOR: Bakkevig K.; Sletta H.; Gimmestad M.; Aune R.; Ertesvag H.;

CORPORATE SOURCE: Degnes K.; Christensen B.E.; Ellingsen T.E.; Valla S. S. Valla, Department of Biotechnology, Norwegian University of Science and Technology (NTNU), N-7491 Trondheim, Norway. svein.valla@biotech.ntnu.no

SOURCE: Journal of Bacteriology, (2005) Vol. 187, No. 24, pp. 8375-8384. .

Refs: 42

ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

046 Environmental Health and Pollution Control

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 12 Jan 2006

Last Updated on STN: 12 Jan 2006

AB Alginate is an industrially widely used polysaccharide produced by brown seaweeds and as an exopolysaccharide by bacteria belonging to the genera *Pseudomonas* and *Azotobacter*. The polymer is composed of the two sugar monomers mannuronic acid and guluronic acid (G), and in all these bacteria the genes encoding 12 of the proteins essential for synthesis of the polymer are clustered in the genome. Interestingly, 1 of the 12 proteins is an alginate lyase (AlgL), which is able to degrade the polymer down to short oligouronides. The reason why this lyase is associated with the biosynthetic complex is not clear, but in this paper we show that the complete lack of AlgL activity in *Pseudomonas fluorescens* in the presence of high levels of alginate synthesis is toxic to the cells. This toxicity increased with the level of alginate synthesis. Furthermore, alginate synthesis became reduced in the absence of AlgL, and the polymers contained much less G residues than in the wild-type polymer. To explain these results and other data previously reported in the literature, we propose that the main biological function of AlgL is to degrade alginates that fail to become exported out of the cell and thereby become stranded in the periplasmic space. At high levels of alginate synthesis in the absence of AlgL, such stranded polymers may accumulate in the periplasm to such an extent that the integrity of the cell is lost, leading to the observed toxic effects. Copyright .COPYRGT. 2005, American Society for Microbiology. All Rights Reserved.

L10 ANSWER 34 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2003405575 EMBASE

TITLE: Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose.

AUTHOR: Spiers A.J.; Bohannon J.; Gehrig S.M.; Rainey P.B.

CORPORATE SOURCE: A.J. Spiers, Department of Plant Sciences, University of Oxford, South Parks Road, Oxford OX1 3RB, United Kingdom. andrew.spiers@plants.ox.ac.uk

SOURCE: Molecular Microbiology, (2003) Vol. 50, No. 1, pp. 15-27. .

Refs: 52

ISSN: 0950-382X CODEN: MOMIEE

COUNTRY: United Kingdom

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 23 Oct 2003

Last Updated on STN: 23 Oct 2003

AB The wrinkly spreader (WS) genotype of *Pseudomonas fluorescens* SBW25 colonizes the air-liquid interface of spatially structured microcosms resulting in formation of a thick biofilm. Its ability to colonize this niche is largely due to overproduction of a

cellulosic polymer, the product of the wss operon. Chemical analysis of the biofilm matrix shows that the cellulosic polymer is partially acetylated cellulose, which is consistent with predictions of gene function based on *in silico* analysis of wss. Both polar and non-polar mutations in the sixth gene of the wss operon (wssF) or adjacent downstream genes (wssGHIJ) generated mutants that overproduce non-acetylated cellulose, thus implicating WssFGHIJ in acetylation of cellulose. WssGHI are homologues of AlgFIJ from *P. aeruginosa*, which together are necessary and sufficient to acetylate alginate polymer. WssF belongs to a newly established Pfam family and is predicted to provide acyl groups to WssGHI. The role of WssJ is unclear, but its similarity to MinD-like proteins suggests a role in polar localization of the acetylation complex. Fluorescent microscopy of Calcofluor-stained biofilms revealed a matrix structure composed of networks of cellulose fibres, sheets and clumped material. Quantitative analyses of biofilm structure showed that acetylation of cellulose is important for effective colonization of the air-liquid interface: mutants identical to WS, but defective in enzymes required for acetylation produced biofilms with altered physical properties. In addition, mutants producing non-acetylated cellulose were unable to spread rapidly across solid surfaces. Inclusion in these assays of a WS mutant with a defect in the GGDEF regulator (WspR) confirmed the requirement for this protein in expression of both acetylated cellulose polymer and bacterial attachment. These results suggest a model in which WspR regulation of cellulose expression and attachment plays a role in the co-ordination of surface colonization.

L10 ANSWER 35 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2003227419 EMBASE

TITLE: The *Pseudomonas fluorescens* AlgG protein, but not its mannuronan C-5-epimerase activity, is needed for alginate polymer formation.

AUTHOR: Gimmestad M.; Sletta H.; Ertesvag H.; Bakkevig K.; Jain S.; Suh S.-J.; Skjak-Braek G.; Ellingsen T.E.; Ohman D.E.; Valla S.

CORPORATE SOURCE: S. Valla, Department of Biotechnology, NTNU Norwegian Univ. Sci./Technol., N-7491 Trondheim, Norway.
svein.valla@biotech.ntnu.no

SOURCE: Journal of Bacteriology, (2003) Vol. 185, No. 12, pp. 3515-3523. .
Refs: 51
ISSN: 0021-9193 CODEN: JOBAAY

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology
029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 26 Jun 2003
Last Updated on STN: 26 Jun 2003

AB Bacterial alginates are produced as 1-4-linked β -D-mannuronan, followed by epimerization of some of the mannuronic acid residues to α -L-guluronic acid. Here we report the isolation of four different epimerization-defective point mutants of the periplasmic *Pseudomonas fluorescens* mannuronan C-5-epimerase AlgG. All mutations affected amino acids conserved among AlgG-epimerases and were clustered in a part of the enzyme also sharing some sequence similarity to a group of secreted epimerases previously reported in *Azotobacter vinelandii*. An algG-deletion mutant was constructed and found to produce predominantly a dimer containing a 4-deoxy-L-erythro-hex-4-enopyranosyluronate residue at the nonreducing end and a mannuronic acid residue at the reducing end. The production of this dimer is the result of the activity of an alginate lyase, AlgL,

whose *in vivo* activity is much more limited in the presence of AlgG. A strain expressing both an epimerase-defective (point mutation) and a wild-type epimerase was constructed and shown to produce two types of alginic molecules: one class being pure manuronan and the other having the wild-type content of guluronic acid residues. This formation of two distinct classes of polymers in a genetically pure cell line can be explained by assuming that AlgG is part of a periplasmic protein complex.

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ACCESSION NUMBER: 2001403406 EMBASE

TITLE: Characterization of algG encoding C5-epimerase in the alginic biosynthetic gene cluster of *Pseudomonas fluorescens*.

AUTHOR: Morea A.; Mathee K.; Franklin M.J.; Giacomini A.; O'Regan M.; Ohman D.E.

CORPORATE SOURCE: D.E. Ohman, Department of Microbiology, M.C.V.C. Virginia Commonwealth Univ., 5-047 Sanger Hall, 1101 E. Marshall Street, Richmond, VA 23298-0678, United States.
deohman@hsc.vcu.edu

SOURCE: Gene, (31 Oct 2001) Vol. 278, No. 1-2, pp. 107-114. .

Refs: 33

ISSN: 0378-1119 CODEN: GENED6

PUBLISHER IDENT.: S 0378-1119(01)00685-0

COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 004 Microbiology

LANGUAGE: English

SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 30 Nov 2001
Last Updated on STN: 30 Nov 2001

AB The organization of the alginic gene cluster in *Pseudomonas fluorescens* was characterized. A bank of genomic DNA from *P. fluorescens* was mobilized to a strain of *Pseudomonas aeruginosa* with a transposon insertion (algJ::Tn501) in the alginic biosynthetic operon that rendered it non-mucoid. Phenotypic complementation in this heterologous host was observed, and a complementing clone containing 32 kb of *P. fluorescens* DNA was obtained. Southern hybridization studies showed that genes involved in alginic biosynthesis (e.g. algD, algG, and algA) were approximately in the same order and position as in *P. aeruginosa*. When the clone was mobilized to a *P. aeruginosa* algG mutant that produced alginic acid polymannuronate due to its C5-epimerase defect, complementation was observed and the alginic acid from the recombinant strain contained L-guluronate as determined by proton nuclear magnetic resonance spectroscopy. A sequence analysis of the *P. fluorescens* DNA containing algG revealed sequences similar to *P. aeruginosa* algG that were also flanked by algE- and algX-like sequences. The predicted AlgG amino acid sequence of *P. fluorescens* was 67% identical (80% similar) to *P. aeruginosa* AlgG and 60% identical (76% similar) to *Azotobacter vinelandii* AlgG. As in *P. aeruginosa*, AlgG from *P. fluorescens* appeared to have a signal sequence that would localize it to the periplasm where AlgG presumably acts as a C5-epimerase at the polymer level. Non-polar algG knockout mutants of *P. fluorescens* were defective in alginic acid production, suggesting a potential role for this protein in polymer formation. .COPYRGT. 2001 Elsevier Science B.V. All rights reserved.

L10 ANSWER 37 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 83218444 EMBASE

DOCUMENT NUMBER: 1983218444

TITLE: Selection of attachment mutants during the continuous culture of *Pseudomonas*

fluorescens and relationship between attachment ability and surface composition.
AUTHOR: Pringle J.H.; Fletcher M.; Ellwood D.C.
CORPORATE SOURCE: Dep. Environ. Sci., Univ. Warwick, Coventry CV4 7AL, United Kingdom
SOURCE: Journal of General Microbiology, (1983) Vol. 129, No. 8, pp. 2557-2469.
CODEN: JGMIAN
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
ENTRY DATE: . Entered STN: 9 Dec 1991
Last Updated on STN: 9 Dec 1991

AB A strain of *Pseudomonas fluorescens* that had been isolated from a freshwater source on a plastic substratum was grown in continuous culture in minimal medium. The 'adsbubble' process (adsorptive bubble separation process) was found to foam-fractionate wild-type cells from the fermenter during flow conditions. This selection pressure favoured the enrichment of two major classes of mutant, both having cell surface characteristics fundamentally different from the wild-type. The wild-type produced very little extracellular polysaccharide, whereas a 'mucoid' mutant, found predominantly in the aqueous-phase, produced an alginate exopolymer. The second class of mutant was isolated from the walls of the fermenter and, like the wild-type, produced little exopolymer. This mutant, with crenated colony morphology, showed increased attachment of solid surfaces compared to the wild-type and mucoid cells when assayed for attachment to polystyrene surfaces for 2 h. Outer-membrane protein, lipopolysaccharides and exopolysaccharides of the wild-type and both mutants were analysed. The results demonstrate the role of cell surface characteristics in the adaptability of the organism to micro-environments such as a solid/liquid or air/liquid interface or the aqueous phase.

L10 ANSWER 38 OF 38 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN
ACCESSION NUMBER: 81225420 EMBASE
DOCUMENT NUMBER: 1981225420
TITLE: Isolation of alginate-producing mutants of *Pseudomonas fluorescens*, *Pseudomonas putida* and *Pseudomonas mendocina*.
AUTHOR: Govan J.R.W.; Fyfe J.A.M.; Jarman T.R.
CORPORATE SOURCE: Dept. Bacteriol., Univ. Edinburgh Med. Sch., Edinburgh EH8 9AG, United Kingdom
SOURCE: Journal of General Microbiology, (1981) Vol. 125, No. 1, pp. 217-220.
CODEN: JGMIAN
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
ENTRY DATE: . Entered STN: 9 Dec 1991
Last Updated on STN: 9 Dec 1991

DATA NOT AVAILABLE FOR THIS ACCESSION N